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VARIETAL SUSCEPTIBILITY OF WHEAT TO TILLETIA LAEVIS KÜHN

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The possibility of the utilization of bunt resistant varieties of wheat as a practical method of control of this disease has attracted the attention of several investigators. Among the hundreds of varieties of wheat which are known, some have been found which possess a marked resistance to bunt, although by far the larger number appear to be susceptible.

The bunt disease is coextensive with the culture of wheat and, in all the important wheat growing regions, is recognized as one of the most serious diseases of this crop. The disease may be caused by one of two closely related species of smuts, *Tilletia tritici* (Bjerk.) Winter and *T. laevis* Kühn. These two species are very similar in their general life history and in their pathological effects upon the host. They vary considerably in their distribution but frequently may be found in the same locality.

Farrer (6) was one of the first investigators to take up the study of the resistance of wheat varieties to bunt. He compared the susceptibility of ten Australian varieties and obtained infections varying from 12.0 to 95.5 per cent. Eight selections from plants of Allora spring wheat, which had escaped infection in the previous year, also were grown. All of these selections gave high infections—87.1 to 95.5 per cent.

Pye (29) compared twenty-one Australian varieties. One of these, Medeah, a Durum wheat, gave negative results. The others were infected to a greater or less extent; some of them quite severely. The varieties Genoa, Florence and Cedar gave the lowest percentages of infection.

McAlpine (20) compared several varieties and hybrids with reference to their susceptibility to *Tilletia laevis*. Federation, Medeah and hybrids of Bobs, Medeah and Tripola parentage, were quite susceptible. Ohio and Genoa proved to be resistant.

[1] Published by permission of the Director of the Agricultural Experiment Station of the University of Missiouri.

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In another report McAlpine (21) discusses more fully some of the results obtained in Australia. The varieties Genoa and Florence were grown at three different stations—Burnley, Dookie and Longerenong—and showed a fair degree of resistance. The average infection at the stations was 14.6 per cent for Genoa and 9.2 per cent for Florence. At Burnley, both *Tilletia tritici* and *T. laevis* were used but no important differences were observed. There also are reported the results with selections of a large number of crosses obtained by Pye at Dookie. Most of these showed high susceptibility but a few with Medeah parentage seemed to possess considerable resistance. A comparison of four varieties, Dexter, Federation, Florence and Genoa, with reference to their behavior to the two species of smut, was made. They responded in a very similar fashion to both.

Darnell-Smith (4) found that Cedar, Florence and Medeah were resistant to both *Tilletia laevis* and *T. tritici*. Comeback, Bobs and Federation gave 100 per cent infection with both species.

Von Tubeuf (39, 40) was one of the first investigators to report results from Europe. In 1900 he compared the behavior of eight varieties to *Tilletia tritici*, two of which, Ohio and Ontario, gave practically negative results and the remaining ones gave infections of 25.4 to 57.6 per cent. In 1901 the variety Ohio again gave practically negative results—Ontario not being grown. The other six varieties gave infections of 62.4 to 77.4 per cent. In this year, sixteen additional varieties also were tested; one, Bestehorns Ueberfluss, proved highly resistant and the others gave infections of 19.7 to 92.7 per cent. Von Tubeuf did not include the partially smutted plants in his data; otherwise his percentages of infection usually would have been higher.

Von Kirchner (16, 17, 18) has made the most extensive studies on the susceptibility of varieties of wheat to *Tilletia tritici*. Altogether he has studied 360 varieties or selections, of which 241 were winter wheats and 119 spring wheats. Most of these varieties belonged to *Triticum vulgare*, but the other species or races of wheat, *Triticum compactum, T. durum, T. dicoccum, T. monococcum, T. polonicum, T. spelta,* and *T. turgidum* were represented by one or more varieties. Many of the varieties were grown two or more years but in large part they were tested only one season. Further, von Kirchner generally recorded only the number of infected heads and did not give his results on the basis of the percentage of plants or heads infected. It also may be noted that great differences were observed in the amount of smut in a given variety in different years.

Von Kirchner found very marked variations in the susceptibility of the varieties. Only a few seemed to possess a high degree of resistance. It is especially interesting to note that one or more varieties of *Triticum compactum, T. turgidum, T. spelta, T. dicoccum,* and *T. vulgare* were very

susceptible. On the other hand, the varieties of *T. durum*, *T. polonicum*, and *T. monococcum* proved to be highly resistant to *Tilletia tritici*, although the infection of one or more varieties of each was obtained.

Malkoff (19) compared the susceptibility to *Tilletia laevis* of some Bulgarian wheats with that of introduced varieties and found that the former were much more susceptible. The Durum varieties were particularly attacked, giving infections of 70.5 to 88.3 per cent. Much lower infections were obtained in 1902–1903 than in 1903–1904.

Appel and Gassner (1) have mentioned that Ohio and Strubes Grannenweizen were highly resistant to *Tilletia tritici*.

Hecke (10) compared five spring wheats with five winter wheats for three seasons with reference to their behavior to *Tilletia tritici*. He found variations in the amount of infection in the different years but as a rule all ten varieties proved to be quite susceptible. On the whole, the highest infections were obtained with the winter varieties. In an experiment on the influence of the date of planting, he compared six varieties and observed great differences in the amount of smut. The variety Ohio, reported by von Tubeuf and Appel and Gassner as resistant, proved to be quite susceptible in Hecke's experiments.

Munerati (24, 25, 26, 27) in Italy has used a number of varieties in his experiments and usually has obtained high percentages of infection. The amount of smut, however, has varied greatly with the environmental conditions and the methods of inoculation. Peglion (28) in Italy compared fourteen strains of twelve varieties and obtained infections of 33.6 to 74.1 per cent. Strampelli (38) also compared fifteen varieties in Italy and obtained infections of 55.0 to 100 per cent. Schribaux (36), in France, found that Manitoba was quite resistant while four other varieties proved to be susceptible. Heuser (11), in his various experiments with *Tilletia tritici*, grew nine different varieties of wheat, all of which were quite susceptible. Various environmental factors were studied and their influence on infection was noted.

In Washington, Gaines (7, 8) inoculated thirteen varieties of wheat with *Tilletia tritici* and obtained percentages of infection from 18.6 to 96.4 per cent. The varieties Turkey and Alaska showed the greatest freedom from infection. In another experiment he obtained 3.4 per cent infection of Turkey, 5.8 per cent of Florence and 70.9 per cent of Hybrid 128. The second and third generations of the cross between Turkey and Hybrid 128 gave infections varying from those comparable to the Turkey variety to others as high as those usually obtained with Hybrid 128. A cross between Turkey and Florence, two resistant varieties, also was studied. Some of the second and third generation families gave much higher infections than either parent.

Stephens and Woolman (37) have recorded their observations on the resistance of a large number of wheat varieties in Oregon to *Tilletia tritici*. Most varieties proved to be highly susceptible, but three, Martin Amber, White Odessa and Red Hussar, showed marked resistance. A large number of selections of Crimean wheats also gave low percentages of infection. No striking cases of resistance were observed among eighty-five Australian and 120 Indian wheats which were grown. Selections from crosses between Turkey and Florence were notably resistant.

Coons (3) has reported some results with *Tilletia laevis* on forty varieties of wheat. Most of these were susceptible, infections of 20 to 66 per cent being obtained. A few varieties, however, showed resistance. These were mainly Turkey or selections from crosses of this type, although one strain of Fultz also is recorded as resistant. Johnston (14) has noted briefly some results in Kansas. He found that hard winter wheats of the Turkey type were resistant to both *Tilletia laevis* and *T. tritici* and that the soft winter varieties were more susceptible to *T. tritici* than to *T. laevis*. The average of twenty varieties of soft winter wheats gave 3.4 per cent infection with *T. laevis* and 25.1 per cent with *T. tritici*.

METHODS AND RESULTS

In the autumn of 1913 the writer began his studies at Columbia, Mo., on the varietal resistance of winter wheats to *Tilletia laevis*. Brief reports (30, 31, 32, 33, 34) on the progress of these experiments have appeared. The present paper is a final statement of the results which were obtained.

The material of *Tilletia laevis* was obtained from a crop of badly infected wheat grown in the vicinity of Columbia. The varieties and selections of winter wheats used were secured from the Department of Farm Crops of the University of Missouri.

On October 2, 1913, the seed of forty varieties of common wheats of Missouri, inoculated with the spores of *Tilletia laevis*, were sown. Practically all of these varieties gave negative results. A few, however, gave very low percentages of infection. Sixty selections from several varieties of common winter wheats also were inoculated. Most of these were sown on October 25 and the remaining ones on November 4. The results are given in table 1.

The chief point of interest in these results with the selections lies in the very high percentages of infection which were secured. By far the larger number gave more than 50 per cent of infected plants and in several the percentage was above 90. These results are in striking contrast to those obtained with the varieties which were sown on the earlier date.

The second point of interest is that no striking cases of resistance were observed. There were differences in the susceptibility of the various selec-

TABLE 1.—*Susceptibility of winter wheat selections to Tilletia laevis Kühn, 1913–1914*

Selection	Total No. of Plants	No. Inf.	Per cent Inf.	Selection	Total No. of Plants	No. Inf.	Per cent Inf.
1	68	56	82.3	43	66	54	81.6
3	91	71	78.0	47	62	51	82.2
4 (X)	90	57	63.3	48	59	50	84.7
4	83	76	91.5	49*	87	68	78.1
5	40	28	70.0	51*	79	40	50.6
7	83	24	28.9	52	80	16	20.0
9	80	57	71.2	53	53	43	81.1
10	60	45	75.0	56*	85	83	97.6
11	78	60	76.9	59	42	34	80.9
12	70	48	68.5	60	49	34	69.3
15	67	62	92.5	61*	61	48	78.6
18 D–4	62	51	82.2	63	48	35	72.9
19	107	96	89.7	64	64	56	87.5
20	62	56	90.3	66*	66	56	84.8
24	77	52	67.5	67	49	33	67.3
25 A–1–5	68	62	91.1	73*	41	25	60.9
26	72	64	88.8	75*	67	58	86.5
28 B–1	74	71	95.9	81	67	58	86.5
28 B–2	74	31	41.8	83*	68	58	85.2
29	80	64	80.0	86	59	51	86.4
29 C–2	58	31	53.4	93	51	30	58.8
30	65	54	83.0	95*	60	36	60.0
32	52	35	67.3	97*	68	46	67.6
33 A–3*	71	56	78.8	98	67	58	86.5
33 A–4	95	74	77.8	101	66	54	81.8
34	46	39	84.7	195	60	57	95.0
35	79	72	91.1	215	45	38	84.4
35 C–1–8	56	44	78.5	245	77	62	80.5
37 (X)	48	33	68.7	247	69	58	84.0
37	65	29	44.6	Black winter Emmer	53	6	11.3
37 A–4	79	71	89.8				
41	70	56	80.0				

* Seed sown Oct. 25, 1913, except the starred numbers which were sown Nov. 4.

tions but none showed any marked degree of resistance to *Tilletia laevis*. Black Winter Emmer, a variety of *Triticum dicoccum*, gave 11.3 per cent infection. All the other selections belonged to *Triticum vulgare*.

Another experiment was carried out in 1913 in which separate lots of seed of three varieties were inoculated with spores of *Tilletia laevis* taken from the same varieties. These results are given in table 2.

TABLE 2.—*Influence of source of spores of Tilletia laevis Kühn, 1913–1914*

Source of Spores	Early Ripe			Red Cross			Treadwell		
	Total No. of Plants	No. Inf.	Per cent Inf.	Total No. of Plants	No. Inf.	Per cent Inf.	Total No. of Plants	No. Inf.	Per cent Inf.
Early Ripe*	191	144	75.3	177	133	75.1	171	111	64.9
Early Ripe**	270	176	65.1	227	168	74.0	229	109	47.5
Red Cross*	193	145	75.1	160	128	80.0	185	87	47.0
Treadwell*	180	158	87.7	196	168	85.7	158	107	67.7

* Seed sown October 14, 1913.

** Seed sown November 4, 1914.

No important differences in the infections by the different spore lots were observed, although the spores from the variety Treadwell gave somewhat higher results on all three varieties. The variety Treadwell was consistently less infected than the other two varieties, the percentage of infection varying from 47 to 67.7 per cent. Early Ripe gave infections of 65.1 to 87.7 per cent and Red Cross, infections of 74 to 85.7 per cent. There were two sowings of Early Ripe inoculated with spores from the same variety. The sowing on November 4 gave slightly lower percentages of infection than that of October 14.

In 1914 seven varieties were inoculated and planted on October 28. The results are given in table 3.

TABLE 3.—*Susceptibility of winter wheat varieties to Tilletia laevis Kühn, 1914–1915*

Variety	Total No. of Plants	Number Infected	Per cent Infected
Early Ripe	96	9	9.3
Fultz	121	12	9.9
Harvest Queen	331	168	50.7
Mealy	71	3	4.2
Mediterranean	84	5	5.9
Poole	58	8	13.7
Rural New Yorker	45	2	4.4

Seed sown October 28, 1914.

In general, very low percentages of infection were obtained. The variety Harvest Queen, however, gave 50.7 per cent infection.

In 1915, fifteen varieties were inoculated. One lot of seed of five of these varieties was sown on October 2 and a second lot of seed on October 30.

Seed of seven additional varieties was sown on October 21 and a second lot on October 30. The remaining three varieties were sown on October 30. The results are given in table 4.

TABLE 4.—*Susceptibility of winter wheat varieties to Tilletia laevis Kühn, 1915–1916*

Variety	Seed sown Oct. 2, 1915			Seed sown Oct. 21, 1915			Seed sown Oct. 30, 1915		
	Total No. of Plants	No. Inf.	Per cent Inf.	Total No. of Plants	No. Inf.	Per cent Inf.	Total No. of Plants	No. Inf.	Per cent Inf.
Defiance	96	8	8.3				159	61	38.3
Early Ripe	87	27	31.0				210	105	50.0
Fulcaster	77	37	48.0				172	108	62.8
Fultz				164	85	51.8	198	86	43.4
Harvest King				146	34	23.9	170	50	28.5
Harvest Queen							306	182	59.4
Lebanon				181	83	45.8	256	130	50.7
Mealy							143	71	49.6
Mediterranean				157	72	45.8	175	71	40.5
Michigan Wonder				161	36	22.3	222	67	31.8
Poole				131	71	53.4	195	101	51.7
Rural New Yorker							173	73	42.2
Treadwell				174	47	27.0	232	92	39.9
Turkey	71	12	16.9				262	92	35.1
Velvet Chaff	74	12	16.2				137	60	43.8

The five varieties sown on October 2 and again on October 30 gave much higher percentages of infection in the later planting as compared with the early. The range of infections of the first planting was 8.3 to 48 per cent and in the second 35.1 to 62.8 per cent. There was no marked difference in the infection of the sowings made October 21 as compared with October 30. All fifteen varieties, however, gave quite high percentages of infection.

In 1916, seventeen varieties were grown. Separate lots of seed were sown on September 23, October 10 and November 3. These results are given in table 5.

In every case the infections were unusually low. While none of the varieties gave entirely negative results, the highest percentage of infection obtained with any variety on any date was 6.7 per cent. No important differences are observable in the seedings of the different dates.

In 1917 about thirty varieties were grown and all the varieties proved to be susceptible, the percentages of infection varying from 5 to 80 per cent.

TABLE 5.—*Susceptibility of winter wheat varieties to Tilletia laevis Kühn, 1916–1917*

Variety	Date of Planting								
	Sept. 23, 1916			Oct. 10, 1916			Nov. 3, 1916		
	Total No. of Plants	No. Inf.	Per cent Inf.	Total No. of Plants	No. Inf.	Per cent Inf.	Total No. of Plants	No. Inf.	Per cent Inf.
Beechwood Hybrid	262	16	6.1	114	4	3.5	89	3	3.3
Defiance	349	6	1.7	481	8	1.6	337	3	.8
Dietz	329	6	1.8	204	5	2.4	178	3	1.6
Early Ripe	270	11	4.0	142	6	4.2	86	2	2.3
Fulcaster	264	3	1.1	159	3	1.8	204	5	2.4
Fultz	242	7	2.8	123	6	4.8	195	5	2.5
Harvest King	339	11	3.2	130	6	4.6	94	4	4.2
Harvest Queen	172	4	2.3	339	5	1.4	218	3	1.3
Lebanon	281	10	3.5	114	1	.8	150	4	2.6
Mealy	257	2	.7	110	1	.9	56	1	1.7
Mediterranean	140	4	2.8	92	4	4.3	110	3	2.7
Michigan Wonder	263	17	6.4	173	11	6.3	141	7	4.9
Poole	170	8	4.7	86	4	4.6	83	3	3.6
Rural New Yorker	49	1	2.0	115	1	.8	75	1	1.3
Treadwell	298	10	3.3	117	3	2.5	185	4	2.1
Turkey	190	12	6.3	344	16	4.6	458	31	6.7
Velvet Chaff	268	3	1.1	193	1	.5	142	1	.7

DISCUSSION

The more commonly grown varieties of winter wheats of Missouri were used in the course of these experiments and all proved to be quite markedly susceptible to *Tilletia laevis* Kühn. In at least one or more seasons a large percentage of infected plants was secured and in some years the number infected was very large. This was particularly true of the sowings with certain varieties in 1913 and in 1915. There was, however, very great variation in the percentages of infection obtained in the different years. In 1913 and 1915 a high percentage of infection was secured; on the other hand, the results were low in 1914 and 1916.

In 1913 and in 1915 great variation occurred in the amount of smut depending upon the date of seeding. In 1913, varieties sown on October 2, gave practically negative results, while the selections sown on October 25 and November 4, gave exceptionally high infections—in many cases over 90 per cent. In the experiment in which the spores from different varieties were used, it was found that the sowing of the variety Early Ripe, of October 14, gave a larger percentage of infected plants than the sowing of November 4. In 1915 the percentages of infection of five varieties sown on

October 2, were 16 to 30 per cent lower than on the same varieties sown on October 30. On the other hand, there were no obvious differences in the amount of smut on the varieties sown on October 21, and again on October 30. The three sowings of 1916 were about three weeks apart but no obvious differences appear in the results for the different dates.

A large number of observations have been made on the relation between the date of planting and the prevalence of bunt of wheat. Kellerman and Swingle (15) and Munerati (24, 25) found that late sowing of winter wheats in the autumn resulted in a more severe infection of bunt than early spring sowing. Hecke (10), Munerati (24, 25), Schribaux (36), Müller and Molz (22) and Heuser (11) obtained higher infections in the early sown spring wheats than in the later, while Müller, Molz and Morgenthaler (23) found no definite correspondence between the date of sowing and the amount of bunt. Volkart (41) found more bunt (63.7 per cent) in winter wheat sown early in the autumn than when sown later (39.3 per cent) and Müller and Molz (22) also obtained similar results. Appl (2) and Heuser (11) found that when a series of sowings of wheat inoculated with the spores of *Tilletia tritici* was made in the autumn, the most severe infections occurred neither in the very early nor the very late seeding. The same observations were made by Heald (9) and Schafer, Gaines and Barbee (35) with *Tilletia tritici* in eastern Washington. The situation there, however, is complicated by the fact that soil infestation is known to occur. Malkoff (19) found no important differences in the amount of *Tilletia laevis* in wheat sown on four different dates from October 10 to November 10.

Several investigators have endeavored to determine the relation of moisture, temperature and other soil factors to the infection of wheat by bunt. Von Tubeuf (39) obtained 55.3 per cent infection by *Tilletia tritici* in wheat planted in sandy soil and watered as compared with 29.3 per cent infection of plants in similar soil but not watered. Volkart (41) accounts for the greater infection of wheat by *Tilletia tritici* in his early planting as compared with the later on the basis of a soil moisture more favorable for the rapid germination of the wheat. Appl (2) concluded that the small differences in the temperature ranges were not sufficient to explain his results with *T. tritici*. From his examination of the records on the amount of precipitation during the period of germination he concluded that soil moisture is of greater importance than temperature for successful infection. Hungerford (12) also concluded that moisture is particularly important in the infection of wheat by *T. tritici* in Idaho where soil infestation occurs.

As regards *Tilletia laevis*, Ivanoff (13) suggests the importance of soil moisture. Malkoff (19) also found different amounts of smut in several varieties of wheat in two successive years. His analysis of the meteorological conditions led him to conclude that the amount of moisture in the soil

during the germination period of the seed had a very marked effect upon the extent of the subsequent infection.

Hecke (10) attempted to correlate the amount of *Tilletia tritici* in spring wheat with the temperature conditions. The highest percentages of infection were obtained in the early sowings although some rather marked exceptions occurred. In general Hecke found that with low temperatures there was a higher percentage of infection. Munerati (26) obtained 1.4–12 per cent infection in four varieties of wheat germinated at 18–20° C. These same varieties germinated at 7–8° C. gave 92–100 per cent infection. More recently Munerati (27) has reported some additional results on the temperature relations of the infection of wheat. He used two varieties, Gentile Rosso and Cologna, and obtained the following results: Seed germinated at 22–25° C. gave 0–1.4 per cent infection; germinated at 10–12° C. gave 13.2–39.3 per cent; germinated at 2–4° C. for forty days gave 0–1.9 per cent; germinated at 2–4° C. for twenty days, then at 22–25° C. gave no infection; germinated at 2–4° C. for twenty days, then at 10–12° C. for seven days and finally at 22–25° C. gave 19.7–24.4 per cent. Heuser (11) secured 1.8–6 per cent infection in three varieties germinated at 16–22° C. and 66.2–94.7 per cent in the same varieties germinated at 6–10° C.

As already emphasized, my own results varied widely in the different years and in the seedings made on different dates. It does not seem possible to definitely correlate these results with any specific temperature or moisture conditions. The air temperature before and after the seeding dates, together with the amount of precipitation for the different years, is recorded in table 6. The data given for the period after seeding varies in the number of days, since the time required for the seedlings to emerge from the soil depends somewhat upon the temperature.

Following the first sowing of 1913 the temperature was above 20° C. and very little rain fell. Practically all of the varieties sown gave negative results. The temperature following the remaining three seedings of 1913 varied from 6–10° C. and in every case a considerable amount of rain fell during the germination period. Quite high percentages of infection were obtained. These results suggest that the low temperature, combined with the moisture, was particularly favorable for infection. In 1914 the temperature was about 13–14° C. during the germination period and no precipitation occurred. The results were comparatively low although 50 per cent of bunt was secured in the variety Harvest Queen. In 1915 the average temperature during the first period of germination was 13–14° C. and during the second period was about 15–16° C. and during the third period was about 17° C. Notably higher percentages of infection were obtained from the second and third sowing. No rain fell during the second and third periods of germination. In 1916 the average temperature during

TABLE 6.—*Temperature and precipitation during the period of seeding of winter wheat at Columbia, Mo., 1913–1916*

Date	10 days Before sowing		After sowing		
	Average Temp.	Total Precip.	No. of Days	Average Temp.	Total Precip.
Oct. 2, 1913	15° C.	.74	6	20.5° C.	.05
Oct. 14, 1913	20° C.	.34	15	9.0° C.	2.04
Oct. 25, 1913	8° C.	1.50	20	6.7° C.	.81
Nov. 4, 1913	5° C.	.56	15	8.5° C.	2.20
Oct. 28, 1914	14° C.	.01	10	13.5° C.	0.0
Oct. 2, 1915	18° C.	.02	12	13.0° C.	.89
Oct. 21, 1915	16° C.	1.11	10	15.5° C.	0.0
Oct. 30, 1915	15° C.	0.0	9	17.0° C.	0.0
Sept. 23, 1916	15° C.	.33	10	16.0° C.	1.99
Oct. 10, 1916	20° C.	T	13	11.0° C.	1.21
Nov. 3, 1916	13° C.	.34	10	13.5° C.	1.28

all three periods of germination was 11–16° C. and relatively high precipitation occurred. In every case, low percentages of infection were secured although the temperatures were certainly favorable, since in previous years under similar temperature conditions a large amount of smut was secured. It is possible that the temperature was too high for favorable infections in the first sowing of 1913 but it seems evident that the temperatures were favorable in the other years.

So far as precipitation is concerned the amount was very much greater in 1913 and 1916 as compared with either 1914 or 1915. In 1913, a year of relatively high precipitation and fairly low temperature, a large percentage of infection occurred. On the other hand, in 1915 at a considerably higher temperature and no precipitation, high infections also were secured.

It seems evident that soil moisture, soil temperature and other possible soil conditions are interdependent factors which determine the occurrence of bunt in wheat. Their interaction determines whether infection will take place and also the severity of the attack. Any one of these factors may be a limiting one in the prevention of infection.

Data have been reported on the relative susceptibility of different varieties of wheat to *Tilletia laevis* and *T. tritici*. Von Kirchner (18) used *T. tritici* in his experiments and found that varieties of *Triticum durum* were comparatively resistant. Malkoff (19) used *T. laevis* and found that durum varieties of wheat were very much more susceptible than varieties of *Triticum vulgare*. McAlpine (21) and Darnell-Smith (4) in Australia

found no obvious differences in the susceptibility of some Australian wheats to the two species of *Tilletia*. Stephens and Woolman (37) compared the behavior of eight varieties to the two species and found no noteworthy differences. Johnston (14) obtained very low infections of the soft winter wheats by *T. laevis*, while higher infections were obtained with *T. tritici*. His results with *T. laevis* are surprising in view of the comparatively high infections obtained by Coons (3) as well as the results reported by the writer.

Various investigators have obtained different results with certain varieties. The Australian investigators in general have reported that Medeah was resistant. McAlpine (20), however, obtained as high as 46.6 per cent infection of this variety by *Tilletia laevis*. Von Tubeuf (39, 40), Appel and Gassner (1) and von Kirchner (18) have emphasized the resistance of the variety Ohio. Hecke (10), however, obtained 61.9 per cent infection of this variety. It is quite probable in the light of the results recently reported by Faris (5) with the covered smut of barley that specialized races of species of Tilletia also may occur and the discrepancies recorded by the different observers may in part be explained upon this basis. Further investigations along this line are particularly important.

It is obvious that experiments to determine the resistance of plants to particular diseases must be repeated over a series of years. If positive results are obtained, evidence is at once secured as to the susceptibility of the variety but negative results or low percentages of infection are not definite criteria as to the resistance of the variety in question. One might conclude from the 1916 results that all the soft winter wheats tested possess a high degree of resistance. As a matter of fact, the records for the years 1913 and 1915 show clearly that they are highly susceptible. If one could assure appropriate conditions for high infection in any given season, then one test might serve to determine the resistance of a variety. Since this can not be done under field conditions, conclusions based on the results of one season can not be accepted as final and the determination of the behavior of the variety over a series of years is essential.

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BACTERIAL SOFT-ROT OF TOMATO

S. A. WINGARD[1]

WITH THREE FIGURES IN THE TEXT

A bacterial soft-rot of tomato fruits is of general occurrence in Virginia and is a factor of considerable economic importance. It is the most common fruit rot in the state and probably causes greater losses on the average than any of the other fruit rots of tomato. The writer's attention was first attracted to the disease during the summer of 1918 when it appeared in destructive form in a series of tomato plats on the experiment station farm at Blacksburg. These plats were sprayed for the control of Septoria leaf-blight and they afforded splendid opportunity for the study of soft-rot.

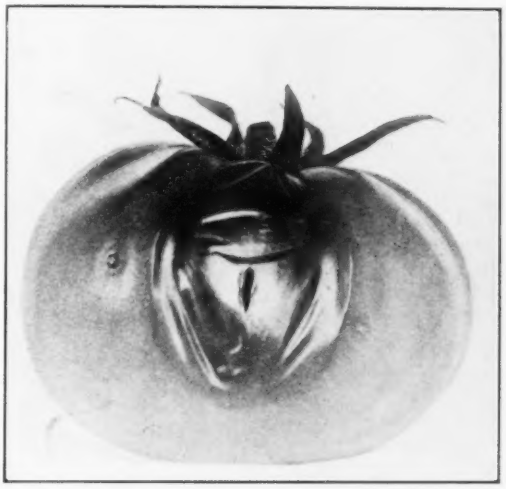

FIG. 1. Tomato fruit 6 days after inoculation with *B. aroideae*. Note the check punctures on either side of the decayed region.

The field studies were supplemented by investigations in the laboratory and greenhouse during the fall and winter months and also during the following year. The results of these studies are briefly described in this paper.

So far as the writer is aware there has been no extended study of the fruit rot in question and practically no information is available in published form. Earle (1) described a bacterial soft-rot of tomato in Alabama in 1900 which he reported as being very destructive. He isolated the bac-

[1] Paper No. 62 from the Department of Plant Pathology, Virginia Agricultural Experiment Station.

terium and proved its pathogenicity by inoculating tomato fruits with pure culture; however, the identity of this organism was not determined. Humbert (3) in 1918 reported a tomato fruit rot as occurring in Ohio. This author referred to the disease as "leak" and stated that bacteria were found associated with it. Sherbakoff (6) referred to tomato soft-rot in a

FIG. 2. Tomato fruits 8 days after inoculation with *B. aroideae*. Note the exudate on the bottom of each.

paper on Florida tomato diseases in 1918, and stated that it was of considerable economic importance in some cases.

DESCRIPTION OF THE DISEASE

Tomato soft-rot is strictly a fruit rot to which the green fruit is especially susceptible. Ripe fruit is not so seriously affected unless infection is initiated while it is still green.

The first indication of infection is the appearance of a water-soaked area on the surface of the fruit (Figure 1). The affected area is depressed with a sharp margin of sound tissue. The tissue within the affected region becomes opaque in a short time, but there is always a narrow band of water-soaked tissue near the margin. The affected region becomes very much wrinkled and often cracks. The entire fruit is converted into a soft, watery, colorless, decayed mass within a period of three to ten days (Figure 2).

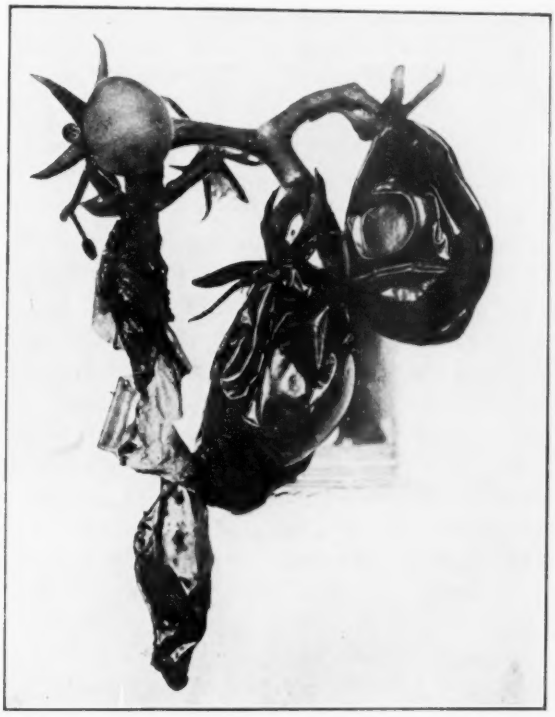

FIG. 3. Tomato fruits 12 days after inoculation with *B. aroideae*. The fruit on the left has burst and "leaked" its contents.

The completely rotted fruits often burst and "spill" their contents as shown in Figure 3. This decay is accompanied by offensive odors.

CAUSAL ORGANISM

The soft-rot organism was readily isolated from the affected tomato fruits on beef-peptone agar. Glistening, grayish-white colonies, which later proved to be those of the soft-rot organism, appeared on this media after a period of 24 hours. A detailed study of the organism was under-

taken by A. B. Massey[2] who has shown that it is referable to *Bacillus aroideae* Townsend[3]. Townsend (7) in 1904 demonstrated that this organism was capable of rotting tomato fruits and certain other vegetables besides causing a rot of the calla lily.

Although the organism in question was found to be *Bacillus aroideae*, it is known that some other organisms are capable of producing similar rots of tomato fruits. Jones (4) in 1900 found that *Bacillus carotovorus* is capable of producing a soft-rot when inoculated into tomato fruits. Pritchard and Porte (5) have shown that a form of *Oospora lactis* produces a watery-rot of tomato fruits which occurs most commonly in shipments of southern grown tomatoes.

PATHOGENICITY

Preliminary inoculations were made on twelve green tomato fruits growing on plants in the greenhouse. These fruits were swabbed with a 1–1000 solution of mercuric chloride and then punctured with the point of a sterile needle which had been previously dipped into a pure culture of the soft-rot organism. Similar green fruits were disinfected and punctured with a sterile needle as controls. Within 24 hours, each of the needle punctures on the inoculated fruits was surrounded by a water-soaked area about 1 cm. in diameter. The decayed spots thus produced were similar in every respect to those which occurred on the fruits in the field. A week later all of the inoculated fruits had been completely transformed into soft-watery masses. The controls remained sound and ripened without any apparent injury. The soft-rot organism was readily reisolated from the inoculated fruits.

In another test, green tomato fruits, which were from 1 to 1.5 inches in diameter, were inoculated by needle puncture and all of them were completely decayed in 11 days.

Another lot of green fruits which ranged in diameter from 1 to 2.5 inches were inoculated in a similar manner. Two of these showed complete decay in 9 days, and four in 12 days. Three others were half rotted in 12 days.

METHOD OF INFECTION

In order to determine whether the soft-rot organism can infect through the unwounded epidermis, green tomato fruits on plants growing in the greenhouse bed were smeared with the organism from pure culture. Other

[2] See A study of *Bacillus aroideae* Townsend, the cause of a soft-rot of tomato, by A. B. Massey in this issue of Phytopathology.

[3] According to the newer classification recommended by the Society of American Bacteriologists the name of this organism becomes *Erwinia aroidea* (Townsend) Comm. S. A. B.

fruits were punctured with a sterile needle and then smeared with a pure culture of the soft-rot organism, and others were punctured and held as controls. The fruits were about one-half mature size. These inoculations were made in the late afternoon in order to allow the organism to remain over night before being exposed to the sun. The results of this experiment are given in table 1.

No infection resulted on any of the unwounded fruits in this experiment, while all of those which were wounded before inoculation showed complete decay within 12 days.

A similar test was made with green fruits removed from the plants and placed in moist chambers at a temperature of 30° C. The fruits which were punctured and inoculated were completely decayed within 7 days, whereas the controls and those which were inoculated without wounding remained sound. The contents of the decayed fruits "leaked" and covered the bottom of the moist chamber to a depth of 5 mm. yet the unwounded fruits, which lay in this bacterial suspension for several days, did not become infected.

TABLE 1.—*Results of inoculation with Bacillus aroideae made on green tomato fruits, on March 3, 1919*

Method of inoculation	Number of fruit	Diameter of decayed spot in cm.	
		March 5	March 12
Organism smeared on punctured surface	1	7	complete decay
	2	9	" "
	3	7	" "
	4	5	" "
	5	5	" "
	6	7	" "
	7	7	" "
Organism smeared on unwounded surface	1	no infection	no infection
	2	" "	" "
	3	" "	" "
	4	" "	" "
	5	" "	" "
	6	" "	" "
	7	" "	" "
	8	" "	" "
Controls, punctured with sterile needle	1	no infection	no infection
	2	" "	" "
	3	" "	" "
	4	" "	" "
	5	" "	" "

Another set of inoculations was made in the greenhouse on 24 fruits of the Norton and Marvel varieties by applying pieces of decayed fruits to their unpunctured surfaces. Within 48 hours, five of these fruits had decayed spots at the stem-end, and one was beginning to decay along a groove

on the side. Within 10 days, fourteen of these fruits were completely decayed, one was partially decayed, and the others remained unaffected. Infection had taken place through the stem ends of twelve fruits, through the blossom ends of two, and through a growth crack in the side of one. It is common to find cracks at the stem and blossom ends of tomato fruits in the greenhouse and it is probable that infection resulted through such cracks. There was no evidence to indicate that the organism had gained entrance through the uninjured skin.

Observations in the field have shown that soft-rot infection takes place through growth cracks, sun scalds and insect injuries.

SUSCEPTIBILITY OF VARIETIES

In order to compare the susceptibility of different varieties to soft-rot, ten fruits of each of the following were inoculated in the greenhouse by needle puncture: Norton, Marvel, Royal Red, Burbank, Red Cherry, and Yellow Pear. The green fruits, which ranged from one-half to two-thirds mature size, were left on the vines. Within 7 days, all of the inoculated fruits were completely or almost completely decayed. All of the varieties were equally susceptible.

SUSCEPTIBILITY OF GREEN AND RIPE FRUITS

Twelve ripe and eight green tomato fruits were punctured and inoculated with the soft-rot organism. Other ripe and green fruits were punctured with a sterile needle and used as controls. All of the inoculated green fruits had rotted completely within 8 days and some of them had dropped from the vines. None of the ripe fruits showed complete decay and only three of them showed slight rotting at the point of inoculation. One of these fruits was completely rotted on the tenth day, while the others, though over-ripe, showed only slight decay at the point of inoculation. The controls remained sound.

A repetition of this test was made with seven ripe and two green fruits inoculated as described above. Two ripe fruits were punctured with a sterile needle for controls. Within 3 days, the green fruits were completely rotted, two of the ripe fruits were partially rotted, three completly rotted, and two remained unaffected.

STEM INOCULATIONS

Stem inoculations were made on the tender tips of six young Norton tomato plants growing in the greenhouse. The inoculum was applied to slits which were made in the tips of the plants with a sterile scalpel. No infection resulted.

Additional stem inoculations were made on two Burbank, two Red Cherry, eight Marvel, six Royal Red, two Norton, and eight Yellow Pear tomato plants. The plants were ten to twelve inches high and the inoculum was introduced into a slit near the tip of each. The plants were watered and left exposed in the greenhouse bed. When examined 3 days later, none of these plants showed infection except the two Royal Red which seemed to be slightly affected. These plants, however, showed no infection when examined a few days later.

Two additional plants of the Royal Red variety were inoculated as described above and placed under bell jars. They appeared to be infected 2 days later, but after 5 days all signs of infection had disappeared.

This evidence indicates that the soft-rot organism is not capable of infecting the stems of the tomato plant.

TEMPERATURE RELATIONS

It had been evident throughout the work that the development of decay by the soft-rot organism was modified by temperature, a longer period being required for complete decay when the greenhouse was cool than when it was warm.

Green tomato fruits of approximately equal size were inoculated by needle puncture and placed in moist chambers each of which was placed in an incubator in which temperatures were maintained as indicated in table 2. Two controls were carried with each lot. The number of days required for complete rotting of all the inoculated fruits varied with the temperature as shown in the table, the most rapid development being at the ranges of 28–34° and 31–34°, and the slowest development at 14–15°. There was no development of rot at 37–42° but the fruits became blackened after 5 days and had a cooked appearance in 7 days. None of the controls showed any rot.

TABLE 2.—*Number of days required for complete rotting of green tomato fruits inoculated with Bacillus aroideae, at the specified temperature ranges*

Temperature range	Number of days
14–15° C.	17
18–22° C.	13
28–34° C.	7
31–34° C.	7
37–42° C.	no rot

OTHER HOSTS

Sweet peppers, cantaloupes, eggplant fruits, and Irish potatoes all proved to be susceptible to decay by the soft-rot organism. When inoculated

by needle puncture and kept in moist chambers at room temperature, sweet peppers were completely decayed in 5 days, eggplant in 4 to 5 days, cantaloupes in 3 days, and Irish potatoes in 7 to 21 days. Townsend (7) has shown that his culture of the organism in question is capable of rotting the corm, petiole, and flower stalk of the calla lily, and the green fruits of tomato, eggplant, cantaloupes, sweet peppers and cucumber. He also reported it as causing rot when inoculated into such vegetables as carrot, potato, turnip, salsify, parsnip, onion, radish, cabbage, and cauliflower.

CONTROL

Data on the control of soft-rot by spraying were obtained in 1918 in an experiment conducted at Blacksburg which has been reported by Fromme (2). Bordeaux mixture of the 5–5–50 formula and soap-Bordeaux (4 pounds bluestone, 3 pounds rosin fish-oil soap, 2 pounds quick lime, 50 gallons water) were applied on two plats and a third plat of the same size served as a check. Five applications of the spray materials were made, the limiting dates being July 6 and August 20. Fruit yields were recorded by weight, the unsound and sound fruits being recorded separately. Practically all unsoundness was due to soft-rot which developed with great severity during an unusually wet period in late August and early September. The total yield for the season from these plats was as follows:

Pounds of fruit	Sound	Unsound
Soap Bordeaux	833	105
Standard Bordeaux	546	207
Check	542	325

The soap-Bordeaux reduced the injury from soft-rot to a marked degree, the percentage control in comparison with the check being 68, while that of standard Bordeaux was 36 per cent. There was no clear explanation for the greater efficiency of the soap-Bordeaux. According to Fromme: "It may have been due to greater bactericidal properties of the spray material, to better covering and adhesion to the fruits, or possibly to foliage protection which decreased sun scald and which would promote more uniform growth of fruit, with fewer growth cracks."

The efficiency of soap-Bordeaux would have been even more pronounced had not the early frost prevented the ripening of the large quantity of green fruit which was left on the plants at the end of the harvest. The soap-Bordeaux plat had 437 pounds of mature green fruit harvested from it after frost and the standard Bordeaux had 424 pounds, while the check yielded only 47 pounds.

SUMMARY

Bacterial soft-rot of tomato, caused by *Bacillus aroideae* Townsend, is capable of causing severe losses in Virginia.

The soft-rot organism is a wound parasite which gains entrance into the tomato fruit chiefly through the growth cracks, sun scalds and insect injuries.

Green tomato fruits are more susceptible to injury from soft-rot than ripe fruits.

All varieties of tomatoes tested were found to be equally susceptible to soft-rot.

The soft-rot organism does not appear to be capable of infecting the stems of the tomato plant.

The optimum temperature for the development of soft-rot appears to be about 30° C.

Losses from soft-rot are appreciably reduced by spraying with soap-Bordeaux.

VIRGINIA AGRICULTURAL EXPERIMENT STATION

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A STUDY OF BACILLUS AROIDEAE, TOWNSEND, THE CAUSE OF A SOFT ROT OF TOMATO, AND B. CAROTOVORUS JONES

A. B. Massey[1]

With Three Figures in the Text

Introduction

In the summer of 1918, at Blacksburg, Virginia, there developed a considerable amount of a soft rot of tomatoes. This occurred in experimental plots which were designated to study the control of septoria leaf blight, and the soft rot of the fruit developed into an important factor. In describing these experiments Fromme (2) states: "Practically all of the unsoundness of the fruit was caused by bacterial soft rot, a disease which is exceedingly common and often very destructive in tomato fields in Virginia." Isolations from diseased fruits made by S. A. Wingard (15) proved a bacterium to be the causative agent. Its growth in pure culture resembled that of the group of bacteria which causes soft rots of plants but it could not be readily assigned to any of the described species of this group. There has been only casual mention of a bacterial soft rot of tomato in literature, and the distinguishing features of the organisms which might be responsible have not been as sharply defined as is desirable. It was decided, therefore, to undertake comparative studies of the organism in question together with some of the non-chromogenic soft rot forms.

Between the years 1898 and 1904 several species of soft rot bacteria were described. The literature of these is summarized by Harding and Morse (4) in their discussion of comparative cultural studies carried out by them. They describe here the situation of the soft rot forms up to 1909. In 1910 Giddings (2) described, under the name of *Baccillus melonis*, a soft rot bacterium found destructive to muskmelons and also capable of producing rot in some other plants.

Harding and Morse (4) seem to be the only ones who have made extensive comparative studies of the typical soft rot forms. Their studies included four named species (*Bacillus carotovorus* Jones,[2] *B. oleraceae* Harri-

[1] Paper No. 63 from the Department of Plant Pathology, Virginia Agricultural Experiment Station.

[2] In accordance to the classification recommended by the Committee of the Society of American Bacteriologists the names used here would be of the genus Erwinia, *Bacillus carotovorus* Jones, becomes *Erwinia carotovora* (Jones) comm. S. A. B., and *Bacillus aroideae* Townsend becomes *Erwinia aroidea* (Townsend) comm. S. A. B. These names are recognized but at this transition period it is found that it is clearer to use the old generic name Bacillus in this paper where synonomy is involved.

son, *B. omnivorus* Van Hall, and *B. aroideae* Towns.) and 39 strains isolated from various soft rot tissues. All of the 43 strains were alike in the 38 classificatory features studied except in their manner of fermenting the common sugars. *Bacillus carotovorus*, *B. oleraceae* and *B. omnivorus*, together with 30 of the unnamed strains composed one group on the basis that they produced acid and gas from dextrose, sucrose and lactose. *Bacillus aroideae* and three of the unnamed strains composed a second group which showed the characteristic of acid fermentation of these three sugars without gas formation. The six other unnamed strains were intermediate between the two groups and varied in the fermentation of these sugars, some producing acid and gas from one sugar and others from two. This shows a difference which it would seem is hardly sufficient basis for the establishment of distinct species. This is further strengthened by the fact that the gas fermenters are weak in their action. The amount of gas usually being just sufficient to recognize its presence in the fermentation tube. Harding and Morse question the advisability of considering *B. carotovorus* and *B. aroideae* as distinct species. However they recognize the fact that their behavior in pathogenicity may separate them more distinctly or show their close relationship.

COMPARATIVE STUDIES

Recognizing the results of the work of Harding and Morse as sifting the usually termed soft rot forms down at least to the two species, *B. carotovorus* Jones, and *B. aroideae* Town. and probably to one (the former), these two forms, along with the strain from tomato, were included in the study. *Bacillus melonis* was not included since a comparison of Gidding's description of this species and Townsend's description of *B. aroideae* fail to show clear specific differences (See table 3). Giddings does not seem to have compared his organism with *B. aroideae* and Erwin F. Smith who has studied the two together states (12, p. 240) "I think that *Bacillus aroideae* and *Bacillus melonis* are identical."

Our culture of *B. carotovorus* was secured from L. R. Jones and is a descendant of his original isolation maintained in his laboratory as No. 3a. The culture of *B. aroideae* was provided by Erwin F. Smith. It is not a descendant of Townsend's original isolation, but one isolated by Smith from calla and believed by him to be the true *B. aroideae*.

The investigations here reported are largely as to parasitism of the organisms in plants of the vegetable and floral groups and also their action towards various organic carbon compounds in pure culture. All inoculations and fermentations were carried out in duplicate and were repeated, with a few exceptions to be noted later. The experiments with the three organisms were carried in parallel and maintained under the same conditions.

PATHOGENICITY

The data here reported are as to inoculations carried on in the field, greenhouse and laboratory. All inoculations were by needle punctures into the uninjured healthy tissue. Potatoes, fruit, etc., in the laboratory were not sliced under aseptic conditions and placed in sterile dishes, but the whole structure was thoroughly cleaned and put into moist chambers and inoculated by puncturing with needle. Moist chambers were used which were large enough to accommodate material for two or more separate inoculations and one or two checks; the latter being punctured with sterile needle. In greenhouse and field, inoculations were made in the same manner into plants and plant parts *in situ*.

Methods of inoculation reported by previous investigators, in studying the soft-rot bacteria, have been of two types. The method which has been followed, as mentioned above, was also used by Jones (7) and is to-day largely used in investigations of wound parasites.

Townsend's method (13) of studying the action of *B. aroideae* was largely that of cutting slices of the plant tissue aseptically, placing in sterile petri dishes, dividing the slice into four parts, orienting them in the dish and finally two pieces in each dish "were inoculated with a 24 hour-old beef broth culture on the surface of the pieces and then stabbing through these drops—with a sterile needle." The objections to this method are twofold. The chances of autolysis of the plant tissue is greatly increased over that of the small wound caused by a needle puncture, hence the bacteria find simpler compounds at hand than those which occur naturally in the tissues. Secondly, the addition of a liquid culture to the wounded surface gives a chance for a saprophyte to appear parasitic since it is possible for the extra cellular enzymes to bring about hydrolysis of the compounds of the tissues. This is nicely illustrated in inoculation of the sweet potato with Rhizopus. Direct inoculations are rarely successful, but infection is more often obtained when the fungus is placed in contact with the wounded surface along with a broth in which the fungus has been growing.

It has been attempted to repeat as far as possible the inoculations reported by others, with these organisms under parallel conditions. Table 1 summarizes inoculations by previous workers, Jones, (7), Smith (12), and Townsend (13), and the results obtained by the author, with the two named organisms and the tomato strain.

The majority of inoculations made in the plants shown in table 1 gave the same results as have been before reported by other workers. As a whole, the negative and positive inoculations indicate a very close relationship between *B. carotovorus* and *B. aroideae*, and show the identity of the tomato strain with *B. aroideae*. They are sharply distinguished, however, by the reaction with certain hosts, notably cauliflower, kohl-rabi, and tobacco. The

TABLE 1.—*Results of inoculations of soft rot bacteria into vegetables, fruits and tobacco. O indicates no soft rot developed; + typical soft rot developed; blank spaces occur where no report has been made, or in author columns, inoculations not made.*

	B. carotovorus			*B. aroideae*		Tomato strain
	Jones	Smith	Author	Townsend	Author	Author
Apple, ripe	O		O	O	O	O
Banana, green			O		O	O
Banana, ripe	O		O	O	O	O
Beet root	O		O		+	+
Cabbage	+		+	+	+	+
Carrot, root	+	+	+	+	+	+
Cauliflower	O		O	+	+	+
Celery	+		+		+	+
Cucumber		+	+	+	+	+
Eggplant fruit	+		+	+	+	+
Kohl-rabi			O		+	+
Lettuce		+	+			
Muskmelon		+	+	+	+	+
Onion, youg leaf	+		O	O	O	O
Onion, mature	+		+	+	+	+
Parsnip	+		+	+	+	+
Pepper, fruit	+		+	+	+	+
Potato, sweet, root	O		O		+	+
Potato, white, tuber	O	+	+	+	+	+
Potato, white, stem	O		O		O	O
Radish	+		+	+	+	+
Salsify	+		+	+	+	+
Tomato, fruit	+		+	+	+	+
Tomato, young stem	O		O	O	O	O
Turnip, root	+		+	+	+	+
Tobacco, stem			O		+	
Tobacco, suckers			O		+	

type of rot produced by the two named organisms is very similar in all cases. Occasionally one finds a slight difference in the color and odor of the disorganized tissues. The results obtained with certain of the hosts are described in the following.

Cauliflower: The writer's inoculations with *B. carotovorus* in the cauliflower were negative agreeing with those of Jones while the inoculations with *B. aroideae* are positive and agree with those of Townsend. It was noticed, however, that the inoculations with *B. carotovorus* developed an initial appearance along a line of punctures as though rot were setting in. However, this initial appearance soon disappeared resulting in no disorganization of the tissue beyond the needle punctures. With *B. aroideae* and the tomato strain this initial appearance developed during the first 24 hours and was followed by rapid discoloration and general breaking down

of the tissues, typical of the soft rot. Harrison (5) reported a soft rot of the cauliflower by his *B. oleraceae,* and Harding and Morse (4), on the basis of their cultural studies, placed Harrison's bacillus in a group with *B. carotovorus.* Jones, as mentioned above, noticed a difference in the enzymatic activity of some soft rot forms. He found Harrison's bacillus to be more active than *B. carotovorus* but not as active as *B. aroideae.* It appears, therefore, that the enzymatic activity of *B. carotovorus* is not such as to enable it to carry out the disorganization of the cauliflower tissue.

Kohl-rabi: A number of inoculations were made into kohl-rabi "balls" both in moist chambers in the laboratory and in the "balls" growing in the

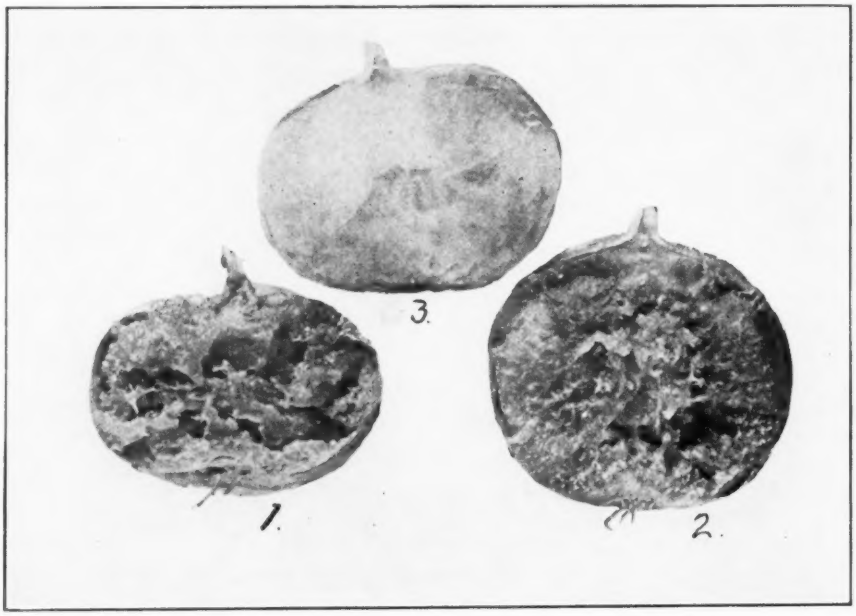

FIG. 1. Soft rot of Kohl-rabi balls induced by inoculation with (1) *Bacillus aroideae* and (2) the tomato strain; (3) Check, punctured with sterile needle.

greenhouse and in the field. The inoculations made with *B. aroideae* and the tomato strain developed rapidly into a mushy rot involving the parenchyma which is enveloped by the fibrous outer layer of the "ball." The rot does not become evident externally for a number of days. A foamy secretion is often noticed from the point of inoculation giving evidence of a considerable gas production. In a firm, succulent "ball" the internal break-down progresses rapidly and in about a week, occasionally in five days, the collapse of the outer wall of the "ball" becomes marked. In figure 1 are shown sections of kohl-rabi "balls" cut three days after inocula-

tion. No. 1 was inoculated with *B. aroideae;* No. 2 with the tomato strain; the upper figure, No. 3, is a check. In these figures are shown the general break-down of the tissues but no effect on the wall of the "ball." The odor of the decayed kohl-rabi is very disagreeable, of the nature of spoiled cabbage but stronger.

The inoculations made with *B. carotovorus* developed initial appearance as described in inoculations of cauliflower with this organism. The change did not go beyond this and even the initial appearance very soon disappeared. Checks which were produced by punctures with sterile needles did not show this change as was the case with the *B. carotovorus* inoculations; hence it could not be considered a case of traumatism.

Onions: Inoculations into mature onion bulbs developed rot in the case of all three organisms. However, the development of the rot by the *B. aroideae* and the tomato strain was slow. Townsend also found rot development in this host to be slow. The inoculations made by *B. carotovorus* developed into a more rapid rot and involved a larger area of the bulb. Only those bulb scales which were punctured with the inoculating needle were affected. The scales beyond the needle puncture become infected rarely and then apparently only through wounds caused by other agencies. Inasmuch as the organism is a wound parasite, this is exactly what might be expected since the bulb of course consists largely of succulent leaf bases. Inoculations into young onions were negative in both cases. Jones reports successful inoculations into the leaf. From his decsription, however, it would seem that the inoculations were made into the leaves of the sprouts from mature onions which were in moist chambers. The young leaves used in inoculations made here were those on young plants growing in soil in the greenhouse.

From this the conclusions to be drawn agree with the accepted facts to be found in the literature of today. Bacterial rot of the onion would not become troublesome in the field until late in the growing season when the bulb is maturing, its most serious damage more often developing in storage or transit and *B. carotovorus* being more actively responsible.

Sweet potato: Jones reports inoculations of *B. carotovorus* into sweet potatoes as unsuccessful and my tests with this organism were also negative. In the first experiments with the sweet potato *B. aroideae* and the tomato strain also proved non-pathogenic. All of these first inoculations were made into roots bought on the market. Later inoculations were made into roots freshly dug from the ground. In these *B. carotovorus* proved non-pathogenic while *B. aroideae* and the tomato strain developed a soft rot involving a small area of the root. The decay caused by *B. aroideae* was not extensive and it is not believed that it would develop a serious soft rot of the sweet potato.

White potato: Inoculations made into the tuber of the white potato from the market or from local storage did not develop pathogenically for *B. carotovorus* but did develop pathogenically in the case of inoculations with *B. aroideae* and the tomato strain. In the latter the decay was rapid and discoloration of the tissues was very marked, being similar to that produced by the blackleg organism. Later inoculations with *B. carotovorus* and *B. aroideae* into tubers recently dug developed into a rot in all cases the same results have been repeated by the writer several times. In these freshly dug tubers the rot by *B. carotovorus* is not essentially different from that produced by *B. aroideae*.

Tobacco: Johnson (6) observed a decay of the tobacco stems in the field which resulted in the development of a hollow stalk. He gives brief mention of the trouble and states that he isolated a bacterium from the diseased tissue with which he was able to reproduce the disease in other plants. He considered the organism one of the soft rot bacteria. More recently reports have come from Connecticut and Massachusetts of isolated cases of the same disease. In these latter cases it seems that no isolations of the organism were made and little or no attempt was made to establish its identity. The writer has made inoculations into tobacco stems and the stem of tobacco suckers with both *B. carotovorus* and *B. aroideae*. Inoculations made with *B. aroideae* developed a rapid browning of the pith followed by a general soft rotting and collapse resulting in a disappearance of the pith of the stem; consequently the "hollow stalk." These inoculations developed very markedly in 36 hours under greenhouse conditions (see figure 3). The inoculation made with *B. carotovorus* did not develop any discoloration or any disorganization of the tissues of the tobacco stalk.

The hollow stalk disease has been described as entering the base of the stalk or the tip and rapidly developing throughout the pith of the stalk and into the petiole and larger veins of the leaf. One plant becoming infected in the field through some wound may easily develop a source for spread of infection by various agencies, especially by the knife in topping. Clinton mentions an isolated occurrence of the disease in Connecticut and states that there are usually two or three plants together in the field. The writer has not found any report of any extensive development of hollow stalk. However, it is possible that the disease has been mistaken for the manifestations of other organisms. Specimens of diseased tobacco plants have been received from Virginia which were difficult to place and it seems probable now that some of these may have been cases of hollow stalk caused by *B. aroideae*. It is possible to confuse hollow stalk and tobacco wilt (*B. solanacearum*) on casual examination.

Striking differences in pathogenicity are also found among the floral plants, especially so in the calla lily and in the wild and cultivated iris. The results of inoculations into floral plants are presented in table 2.

TABLE 2.—*Results of inoculations of floral plants with soft rot bacteria.*

	B. carotovorus	*B. aroideae*	Tomato strain
Common Calla Lily, petiole *Zantedeschia aethiopica.* Var. *Minor*	O	+	+
Golden Yellow Calla, petiole *Zantedeschia Elliotiana*	O	+	+
Elephant Ear, petiole *Caladium esculentum*	O	O	O
Garden Gladiolus, green leaf	O	O	O
Blue Flag *Iris versicolor* Leaf	+	O	O
Wild Spring Iris *Iris verna* Leaf	+	O	O
Hyacinth, Scape	+	+	+

As is noted in the table none of the organisms infected the elephants' ear nor the gladiolus, and all proved positive for the hyacinth. The action towards calla and iris is especially striking and is detailed as follows:

Calla lilies: Toward this host *B. aroideae* and the tomato strain are markedly pathogenic while *B. carotovorus* is not at all so. This is the host from which Townsend first isolated his organism. The usual illustration of the effect of this organism on cells shows the breaking down of the tissues over a limited area of the petiole. Such is not the most striking manifestation of the disease. The limited area rapidly enlarges resulting in a general destruction of the host as shown in figure 2. Leaf blade inoculation likewise results in a disorganization of the tissues. Inoculations from vigorous cultures into a good healthy petiole will show infection within 24 hours while those made into old petioles are slow in development. The writer has found this host to be the most satisfactory one for proving out *Bacillus aroideae.*

Iris: Inoculations into this host gave opposite results from the calla inoculations. *B. carotovorus* proved pathogenic while *B. aroideae* and the tomato strain did not. These inoculations were made mostly into the green leaf under very humid conditions. Infection is apparent as a general breaking down of the leaf tissues in a circular fashion around the needle puncture. Diseased areas on leaves which develop during periods of high humidity dry up and fall out when the humidity decreases, giving the appearance of shot hole, or if they occur on the edge of the leaf, resemble more the depredations of insects.

Observations for several years in a local garden where soft-rot of tomato has been destructive are of interest. In this garden soft rot of carrots, white potatoes, parsnips, cucumber, muskmelon and other plants was common while no rhizome or leaf rotting of iris has appeared. Irises taken from this garden proved by inoculations to be susceptible to *B. carotovorus*.

FIG. 2. Calla lily plants inoculated in petiole (1) with *B. carotovorus*, (2) with *B. aroideae*. No infection results from *B. cartovorus* inoculations.

Iris has been reported as subject to bacterial soft rot by several workers and it is not an uncommon disease. Specimens have been received from two localities in Virginia. Van Hall (14) described a disease of the young shoots caused by his *Bacillus omnivorus* (which according to the work of Harding and More mentioned at the outset, comes to *B. carotovorus*) on *Iris florentina* and *I. germanica*. Richardson (10) reports *B. carotovorus* as the cause of soft rot of iris in Canada. Shull (11), in a popular article, described the disease and measures of control.

Miss Lacy (9), in a short note, reports a soft rot of violet caused by *B. carotovorus*. So far our inoculations have all been negative. Difference

in varieties and conditions under which grown most likely is the explanation. The plants used in our work were cultivated forms that received no care. Miss Lacy's plants were from a garden where they were attended to and the growth was very likely much more succulent than was the case of the plants used in this experiment. Miss Lacy gives a very brief description of the organism she isolated and found to be parasitic and mentioned

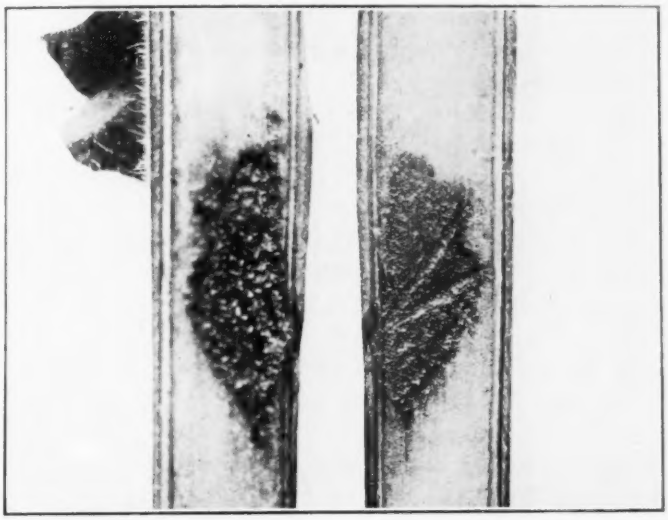

FIG. 3. Longitudinal section of tobacco stem 36 hours after inoculation with *B. aroideae*, showing disintegration of the pith.

that it corresponded favorably with *B. carotovorus* in culture in her laboratory but does not state that the pathogenicity of the latter organism was determined.

FERMENTATION

Cultural differentiation of *B. carotovorus* and *B. aroideae* has been mainly based upon the fact that the former produces gas where the latter does not. Bearing in mind the small amount of gas produced by *B. carotovorus* it makes the cultural differentiation rather uncertain.

The action of the organisms toward various organic carbon compounds were studied for the most part in solution. The cultures were grown in 150 cc. Erlenmeyer flasks, each containing 25 cc. of the culture solution. The solution was prepared by dissolving the total amount of organic carbon compound in four-fifths of the total amount of distilled water to be used. The concentrations of the organic compounds used are shown in table 4. After the solution was all dissolved 20 cc. portions were measured with

pipette into the flasks. These were plugged with cotton wool and sterilized in the autoclave under 15 pounds steam pressure for 15 minutes. Thus the carbon compounds were sterilized in the presence of the distilled water only. To each of these flasks was next added 5 cc. of sterile 2.5 per cent Bacto peptone solution. The transfer to the flasks was accomplished by means of sterile pipettes under guarded conditions to prevent contamination from the air. This made a peptone concentration of 0.5 per cent in each flask. After the sterile peptone solution had been added the flasks were incubated for three days to develop any contaminations. The percentage of contaminated flasks was very small.

The flasks were inoculated from 24-hour peptone broth cultures by putting into the flasks 2–3 drops of the culture from a pipette. The inoculated flasks were incubated at 25°–30° C. The hydrogen-ion concentration was determined at intervals to note the development of acidity and final pH. These determinations were made colorimetrically by comparing with standard buffer solutions containing the proper indicator. For detecting gas fermentation the agar shake method was used. The media was prepared in the same manner as described for the liquid cultures, *i.e.* by sterilizing the component parts separately. At first the oxygen relation in such cultures was questioned but experiments as shown in tables 5 and 6 cleared this point.

Table 3 summarizes the action of *B. carotovorus*, *B. aroideae* and *B. melonis*, as reported by Jones (7), Townsend, (13) and Giddings (3), respectively, and by Smith (12) for *B. carotovorus*, on some of the common organic compounds used in culture studies.

As shown in table 3 the three organisms are not distinguished on the basis of acid production, but *B. carotovorus* differs from the others in gas production. The amount of gas is often slight and this has not been considered a sufficient basis for separation by Harding and Morse (4).

The table shows how impossible it would be to separate Giddings's bacillus from *B. aroideae* in such cultures.

The fermentations here reported were carried out to determine the actions of *B. carotovorus* and *B. aroideae* toward a larger number of organic compounds in hope of showing stronger their relationship. This was carried out by observing the acid and gas fermentation of the following:

Alcohols

Monohydric—Ethyl, Butyl,
Polyhydric—Glycerol, Mannitol,
Aldehyde—Vanillin

TABLE 3.—*Gas and acid fermentation by B. carotovorus, B. aroideae, and B. melonis as gathered from literature.*

	B. carotovorus				*B. aroideae*		*B. melonis*	
	Gas		Acid		Gas	Acid	Gas	Acid
	LRJ	EFS	LRJ	EFS	Townsend		Giddings	
Dextrose	+	+	+	+	O	+	O	+
Lactose	+	+	+	+	O	+	O	+
Sucrose	+	+	+	+	O	+	O	+
Maltose					O	+	O	+*
Mannitol	+	+	+	+	O	+	O	+
Glycerol	O	O	+	+	O	+	O	+
Muscle sugar		+		+				
Milk	O	O	+	+	+	+**	+	+

* Giddings's results indicate acid development from maltose at 25° C. during first four days to be about pH 6.8, on 18th day about pH 7.4. Townsend's determinations were in twenty-weeks'-old cultures using litmus indicator.

** Reported by Smith (4). Townsend did not test the action of his organism on milk in fermentation tubes.

Monosaccharides

Pentoses—Arabinose, Xylose,
Methylpentose—Rhamnose,
Hexoses—Dextrose, Galactose, Levulose

Disaccharides—Saccharose, Maltose, Lactose,
Trisaccharides—Raffinose,
Polysaccharides—Starch, Dextrin,
Glucosides—Amygdalin, Esculin, Salicin, and Arbutin.

This selection presents a series of organic carbon compounds of varying complexities and was made with the hope of bringing out differences in the organisms through the selective action of their enzymes, that is, through the relationship between molecular configuration of the compounds and enzymatic action.

In the beginning it was realized that by observing only the acid and gas fermentations, the chances of finding differences in the action of the organism were limited. However, for practical differentiation of the organisms in culture a more detailed chemical study of the fermentation, as to the kind and amount of cleavage products, though interesting, and valuable, would not necessarily aid in readily distinguishing the organisms in cultures. It is hoped later to go into the study of these cleavage products which should help to explain points that come to mind in this study, and by chance throw

some light on the cause of resistance and susceptibility in plants to these bacteria.

The results of the fermentation of the compounds listed above, exclusive of the glucosides, are shown in Table 4. The hydrolysis of the glucosides is given in table 5. The relationship of the action on the mono-, di- and trisaccharides (exclusive of maltose) is the same as that shown by previous studies in the fermentation of the usual laboratory sugars. That is, *B. carotovorus* produces acid and gas, while *B. aroideae* and the tomato strain develop acid fermentation only.

The action towards maltose is contrary to previous reports. The writer found a slight rise in the H-ion concentration during the first 48 hours, but the action soon reverses. The figures in parenthesis opposite maltose is the pH. reading at the end of 24 hours. The most feasible explanation of this small acid development is that there seemed to be a trace, not readily detected, of inverted maltose in the culture. This was fermented with development of slight acidity and when used up, maltose proving unfavorable, the protein molecules are attacked to satisfy the carbon as well as the nitrogen requirements in the metabolism of the bacteria. The excess nitrogen being eliminated in the form of ammonia would increase the hydroxyl concentration. An analogous action towards the polysaccharides was found, though no initial rise in the pH.

Bio-chemical studies of the enzymes have shown that *emulsin* is commonly responsible for the hydrolysis of glucosides of the beta type, while the alpha-glucosides are hydrolized by the enzyme *maltase*. The beta-glucosides, amygdalin, aesculin, salicin, and arbutin were hydrolized by both organisms which indicates the presence of the emulsin complex in the cultures of these organisms. Maltose, glucose alpha-glucoside, is not hydrolized by these bacteria which indicates the absence of *maltase*. Demonstrating the presence of emulsin helps in further interpretation of table 4. Lactose, a glucose beta-galactoside, is more actively hydrolized by lactase. Some preparation of the emulsin complex from almonds has been found capable of hydrolizing lactose. The tri-saccharide, raffinose, is hydrolized by both emulsin and invertase; emulsin converting it to sucrose and galactose and invertase to fructose and melibiose. Melibiose (glucose beta-galactoside) is slowly acted upon by emulsin. Finding evidence of the presence of emulsin and invertase in cultures of these bacteria give a clearer understanding of the action shown towards the di- and tri-saccharides of table 4. Through the enzymatic action these compounds (exclusive of maltose) are hydrolized to the simpler monosaccharides and these are in turn fermented forming acid on the one hand and acid and gas on the other. It is in the fermentation of the monosaccharides that a careful biochemical study would be of interest and likely instructive as to the physiology of these closely related forms.

TABLE 4.—*Acid and gas fermentations by B. carotovorus and B. aroideae.*

		B. carotovorus			*B. aroideae**		
		H-ion concentration		Gas	H-ion concentration		Gas
		At start	4-day culture	7 days	At start	4-day culture	7 days
Alcohols and Aldehyde							
Ethyl	4.0 %	6.8	5.2	—	6.8	7.4	—
Butyl	0.5 %	6.8	7.0	—	6.8	7.2	—
Glycerol	6.0 %	6.8	6.8	—	6.8	7.0	—
Mannitol	1.0 %	6.8	5.0	+	6.8	5.0	—
Vanillin	0.02%	6.6	6.2	—	6.6	6.4	—
Monosaccharides							
Arabinose	1.0 %	7.2	5.6	+	7.2	5.6	—
Xylose	1.0 %	6.6	5.6	+	6.6	5.6	—
Rhamnose	1.0 %	7.0	5.2	+	7.0	5.4	—
Dextrose	1.0 %	6.6	5.0	+	6.6	4.4	—
Galactose	1.0 %	6.2	5.2	+	6.2	5.4	—
Levulose	1.0 %	6.6	5.2	+	6.6	5.4	—
Di- and Tri-Saccharides							
Sucrose	1%	6.8	5.0	+	6.8	5.0	—
Maltose	1%	6.8(6.2)	7.8	—	6.8(6.4)	7.2	—
Lactose	1%	6.4	5.2	+	6.4	5.4	—
Raffinose	1%	7.2	5.4	+	7.2	5.2	—
Polysaccharides							
Starch	1%	7.2	7.6	—	7.2	8.0	—
Inulin	1%	7.2	8.0	—	7.2	8.2	—

* The tomato strain agreed with the *B. aroideae* throughout.

The action on ethyl alcohol is of interest as it gives an additional means of cultural differentiation. The acid development from ethyl alcohol is very marked with *B. carotovorus* while it is entirely absent in cultures of *B. aroideae*. Growth in peptone broth containing ethyl alcohol is very striking. *B. carotovorus* produces a heavy pelicle and an abundant growth, while *B. aroideae* produces a very slight growth. This is well illustrated by Smith (12, fig. 184). The writer is not aware of acid fermentation of alcohol by *B. carotovorus* being previously recorded. By use of an alcohol-peptone, Brom-cresol-purple agar the two forms are readily separated as *B. carotovorus* quickly develops acid which changes the indicator from purple to yellow. *B. aroideae* grows on this agar, but there is no change in the color of the indicator.

The method of preparing the alcohol medium is as follows: The agar base contains 1 per cent peptone, 1.8 per cent agar shreds, and Brom-cresol-purple in distilled water. This is dissolved, tubed (8 cc. per tube) and sterilized. Just before use the agar is melted, cooled to 45° C. and 0.4 cc. of 95 per cent ethyl alcohol is added. The tube is rolled to mix and is quickly cooled as a slant. This can be held to prove its sterility if desired. The concentration of alcohol is 5 per cent. Ten per cent alcoholic concentration is not prohibitive to *B. carotovorus*, but seems to be to *B. aroideae* though the tolerance of the forms to alcohol has not received the writer's attention.

Stroke culture on agar of this type reveals acid development very prettily. For isolating *B. carotovorus* from rotted tissue it is of course essential that the plates be poured when the agar is first made on account of the volatility of the alcohol. On these plates *B. carotovorus* is readily isolated by fishing the acid developing colonies. If the rot is due to *B. aroideae* fishing from amoeboid non-acid colonies will assist in isolation.

The writer has also studied authentic cultures of *B. phytophthorus* and *B. atrosepticus* on alcohol-agar and both show no acid or gas production. They disagree with *B. carotovorus* in this respect although they are like it in a number of other respects according to facts gathered from literature and unpublished studies of the writer.

OXYGEN REQUIREMENTS

The growth of the soft rot forms in the closed arm of Smith fermentation tubes brought to mind the question of oxygen requirements. This was tested out in the presence of dextrose only. Two experiments were set up: The pH development was studied, first, in flasks and tubes presenting different surface areas exposed to the air of the vessel and second, in tubes with sterile vaseline covering the surface of the medium.

Experiment No. 1. pH development in dextrose peptone broth under varying surface area exposures. A dextrose peptone broth (peptone 0.5 per cent, dextrose 1 per cent) was prepared in bulk and then distributed in 25 cc. quantities in small (150 cc.) Erl. flasks 6.5 cm. in diameter at the surface of the liquid and large test tubes of 2.5 cm. diameter. After sterilization six of each were inoculated and each day for three days two cultures of each bacterium were used to determine the pH. Just prior to making pH determination in a culture the broth was thoroughly mixed and then determinations were made colorimetrically. The results are given in table 5.

Bacterium angulatum (Fromme & Murray) and *Bacterium tabacum* (Wolf & Foster) are included to show more strikingly the effect of surface area exposed on H-ion development. The tobacco bacteria are strict aerobes;

they do not grow in closed arm of Smith fermentation tube. This demonstrates the facultative nature of the soft rot forms. The availability of free oxygen is not a factor in the fermentation of the carbohydrates by them. The case is different with the tobacco leaf spot organisms. Also it is demonstrated clearly that it is necessary to consider the surface area of the culture in studies of H-ion development unless preliminary experiments show the organism to be a facultative form.

TABLE 5.—*Development of H-ion concentration in dextrose broth with varying surface area exposure.*

Bacteria studied	24-hour culture		48-hour culture		72-hour culture	
	Square centimeters in exposed surface					
	44.18	5.03	44.18	5.03	44.18	5.03
	pH	pH	pH	pH	pH	pH
B. carotovorus	5.0	5.0	5.0	4.8	4.8	4.8
B. phytophorus	5.2	5.2	5.0	5.0	5.8	4.8
B. aroideae	5.0	5.0	5.0	4.8	4.8	4.8
Bact. tabacum	5.2	7.0	5.0	6.4	4.8	6.6
Bact. angulatum	5.6	7.0	5.0	5.4	4.8	5.0
Control	6.6	6.6	6.6	6.6	6.6	6.6

Experiment No. 2. The pH development from dextrose in culture tubes with sterile vaseline covering the medium (Brown's (1) vaseline tube). The medium used here was the same as in the previous experiment; 10 cc. were placed in test tubes 1.5 cm. in diameter and then sterilized in the autoclave. As soon as the autoclave could be opened the tubes were taken out and about 1 cc. of melted sterile white vaseline was placed in each tube. In this way the chances of oxygen being absorbed were slight, if any. After cooling these were inoculated from agar cultures. The inoculation was made direct into the medium by holding the tube at about 45° angle and gently melting the edge of the vaseline which slips, thereby exposing the medium. After inoculation the vaseline plug is melted by holding the tube in the flame, tube placed upright, and the vaseline solidifies over the surface of the medium. That this vaseline seal is effective is shown by the fact that 9 tubes of broth prepared in this way, but not inoculated, standing for eleven months in the open laboratory, with only the protection of the cotton plug, show less than 1 mm. shrinkage.

The results of these anaerobic cultures are shown in table 6.

SUMMARY

A bacterial soft rot of tomato is here shown to be caused by *Bacillus aroideae* Townsend.

TABLE 6.—*Development of H-ion concentration and gas in vaseline sealed tubes in presence of dextrose.*

	pH	5-day-old culture	Gas
B. carotovorus	4.8		+
B. aroideae	4.8		O
B. phytophthorus	4.8		+
Bact. tabacum	6.6	No growth	O
Bact. angulatum	6.6		O
Control	6.6		O

Comparative studies of *B. aroideae* and *B. carotovorus* Jones, indicate the close relationship of these two forms, but they may be readily differentiated by either laboratory cultures, pathogenicity, or, better, by a combination of the two.

Bacillus carotovorus and *Bacillus aroideae*, though closely related, should be maintained as separate species.

The following scheme summarizes the differentiation of the two organisms:

I CULTURAL AND FERMENTATION CHARACTERS

	B. carotovorus	*B. aroideae*
Agar colonies in thinly sown plates	round, entire	amoeboid
Fermentation of: dextrose, lactose, galactose, saccharose, mannitol, etc.	acid and gas	acid without gas
Action in ethyl alcohol media	acid without gas, heavy pellicle and abundant growth.	no acid or gas, no pellicle and slight growth.

II PATHOGENESIS

Inoculation into:		
Calla	negative	positive
Iris	positive	negative
Kohl-rabi	negative	positive
Cauliflower	negative	positive

Studies of the oxygen requirements demonstrate the facultative nature of the soft rot forms and bring out the importance of stating the area of

the exposed surface of a broth in studies of H-ion concentration if the organism is not a facultative type.

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EXPERIMENTS IN CONTROL OF CANKERS OF PEAR BLIGHT

L. H. Day

With One Figure in the Text

The term canker as herein used in relation to the pear blight disease (*Bacillus Amylovorus*) refers to the diseased area of the bark in larger branches. These areas or cankers may vary in size from a fraction of an inch in diameter up to areas surrounding the whole branch or trunk and may extend along such parts for many feet.

It is a quite current opinion held by pear growers and many plant pathologists who have not taken particular notice of the literature on the subject or of the lesions of the disease in the bark, that the disease is primarily in the cambium layer. In fact this statement is quite common in texts, bulletins and articles on pear blight. A number of authors, however, have pointed out the fact that this is not the case, and that the disease often kills only the outer bark. In California where the Bartlett pear industry has usually been very profitable, growers have gone to great expense in fighting blight and in attempts to save larger branches which have become infected. For many years the more progressive growers made a practice of going over their orchards every week or ten days and cutting out the affected areas of bark (cankers) in larger branches before the disease had a chance to progress all the way around. The bark was cut to the wood and some disinfectant applied. Finally, about 1916, one grower, Mr. Hayward Reed, of Sacramento, having noticed that the disease progressed in the outer bark for several weeks before it penetrated inwardly to the cambium layer, tried on a rather large scale, the idea of shaving off only the outer bark over the cankers and applying disinfectants. This method gave considerable promise. Many of the cankers were arrested and the uninjured cambium regenerated a new bark within the season. This method was taken up by a number of growers and gained considerable favor until in the virulent outbreak in 1920–21 it failed completely. At this juncture the writer began an investigation of the matter to determine the possibilities of the method and to try to perfect it, if it seemed feasible.

Hundreds of cankers were scarified, that is, the outer bark shaved off, and treated with various disinfectants. It became evident that there was considerable promise in the method. Reimer's formula consisting of cyanide of mercury and bichloride of mercury, one part each in 500 parts of water, by weight, was the most commonly used disinfectant. However, it was not penetrating enough. Apparently the water evaporated too quickly and left the chemical dry and inactive on the scarified bark. In the spring of 1922, I had a student scarify some thirty-five or forty cankers, to half of which

was applied the Reimer formula in water and to the other half the same disinfectants dissolved, in the same proportions, in a solution consisting of one part water and three parts of glycerine. This latter formula arrested

FIG. 1. Pear blight canker scarified from near the ground and a short distance up each scaffold. The cambium was saved under this canker and a new bark generated.

all the cankers treated while the other gave only thirty-five per cent control. This method and formula is now used by hundreds of pear growers in Cali-

fornia. Those who understand the method of scarifying and have careful workmen secure a high percentage of control, possibly from 80 up to 95 per cent. The older that cankers become the deper they penetrate and the more difficult to arrest. The grower goes over the orchard every week or ten days to find these cankers before the disease becomes too deep.

The method of operating is as essential as the disinfectant formula used. The dead outer bark must be shaved down until most of the diseased tissue is removed. Thick bark in crotches, and around branch collars and old bud scars must be pared nearly to the cambium. However, if pared too deeply the disinfectant will kill the cambium. Not the least speck of epidermal bark should be left in the diseased area for the disinfectant will not penetrate the waxy covering of the bark. The bark must be shaved for four or five inches beyond the ends of the canker. These various details of course can only be learned by experience.

A long bladed jack-knife is a good tool for shaving the bark but the growers have improvised various "scrapers" for this work. The photograph shows a pear tree scarified from near the ground and up each scaffold for a distance of about one foot. It required about one hour to perform this operation.

The author has tried to accomplish these same results with penetrating and caustic chemicals painted onto the bark to take the place of the more laborious surgical operation. Scores of different chemicals have been tried. Some have given promise but there are a number of factors which make this a difficult problem. Branches differ in the readiness with which they take in chemicals. Trees in different orchards vary in this respect, and they vary from season to season in a rather large range. The diseased area usually absorbs chemicals more readily than does the healthy tissues at the margins of the canker, especially in older cankers. The range between a concentration of chemicals sufficient to kill the bacteria and one that will not overpenetrate and injure the cambium is too close to make any of the materials yet tried very satisfactory. The iodine salts have been the most promising, especially zinc iodine and potassium tri-iodide. A fifteen per cent solution of zinc iodide has stopped practically all cankers treated but it over-penetrates so frequently in the older cankers, that many branches are lost that could have been saved by scarification.

Then, of course, varieties of pear trees differ in the readiness with which the bark absorbs chemicals and also the blight runs deeper in some varieties than in others. Possibly in some climates the blight may run deeper than in others so that methods that may be successful in California may not be successful in other places.

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